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# **EXHIBIT A**



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# (12) United States Patent

# Petzelbauer

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# (54) THERAPEUTIC FIBRIN-DERIVED PEPTIDES AND USES THEREOF

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See application file for complete search history.

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# (57) ABSTRACT

The invention relates to peptides having the general formula (I), or a salt or amide thereof, wherein  $R_1$  and  $R_2$  are either the same or different, wherein  $R_1$  and  $R_2$  are each selected from the group consisting of hydrogen and a saturated or unsaturated hydrocarbon residue, said residue having from 1 to 10 carbon atoms, wherein  $Z_1$  is selected from the group consisting of histidine and proline, wherein  $Z_2$  is selected from the group consisting of an arginine and a peptide comprising an initial arginine and having from 2 to 30 amino acids. The invention also relates to methods using the peptides of the present invention in the treatment of inflammation

$$\begin{array}{c|cccc}
R_1 & H & O \\
N & C & C & Z_1 - Z_2.
\end{array}$$
(I)

# 4 Claims, No Drawings

# THERAPEUTIC FIBRIN-DERIVED PEPTIDES AND USES THEREOF

### CROSS REFERENCE TO RELATED APPLICATION(S)

The present application is a continuation of International Patent Application No. PCT/AT01/00387, filed Dec. 7, 2001, published in German on Jun. 20, 2002 as International Patent Publication No. WO02/248180, which claims priority 10 to Austrian Application No. AT A 2063/2000, filed Dec. 12, 2000, all of which are incorporated in their entireties herein.

The invention concerns peptides and/or proteins, their use for preparing a therapeutic and/or preventive pharmaceutical composition as well as a pharmaceutical composition.

Substances for the inhibition or prevention of inflammatory reactions, so-called immunosuppressants, which so far have been used for prophylaxis and therapy, generally comprise two distinct groups. Firstly, derivatives of a horsecondly, exogenous immunosuppressants cyclosporin and its derivatives, azathioprine, cyclophosphamide etc. All those substances possess anti-inflammatory effects but they show substantial side reactions in long-term therapy. Those side reactions have a limiting effect on 25 long-term therapy, which is why those substances are used alternately or in combination in order to keep side effects on a tolerable level or in order to be able to actually proceed with the therapy. As examples of side reactions, the pathological fractures associated with cortison are to be men- 30 tioned, which fractures are caused by the osteoporotic effect of the cortisone, or the renal failure which may be caused by cyclosporin. Those side reactions are inevitable with both groups of compounds, and hence it is merely a question of the duration of the therapy and of the total dose at what point 35 the therapy must be stopped.

The present invention has as its object to provide new pharmaceutical products which are suitable for preventing or inhibiting inflammatory effects and which only show minor side effects. A further object consists in providing long-term 40 therapy.

In the following, the amino acids of the peptides according to the invention are referred to by the usual abbreviations, which denote the  $\alpha$ -amino acids.

By "analogues", a peptide is understood which, by deri- 45 vatisation, substitution, preferably homologous substitution, deletion and/or insertion, is derived from the sequence of the fibrin and in particular from the preferred sequences.

The peptides or protein according to the invention exhibit the general formula I

$$\begin{array}{c|c} R_1 & H & O \\ & & \parallel & \parallel \\ N-C-C-C-Z_1-Z_2 \\ & \parallel & \parallel \end{array}$$

wherein R<sub>1</sub> and R<sub>2</sub>, being equal or different, denote hydrogen, a saturated or unsaturated hydrocarbon residue comprising from 1 to 3, in particular up to 10, carbon atoms,

 $Z_1$  denotes a histidine or proline residue,

Z<sub>2</sub> denotes an arginine residue, a peptide residue or a protein residue

comprising an initial arginine residue, in particular comprising from 2 to 30 amino acids, as well as the salts thereof, 2

and, f.i., also amides, or mixtures with each other and/or with at least one further substance for therapeutic and/or preventive use in human and/or veterinary medicine, whereby in particular only L-amino acids are provided. Sequences of formula I are listed in Table 1.

It was completely surprising that the specified amino acid sequence prevents the adhesion of cells from the bloodstream to endothelial cells of the vascular wall and/or their subsequent transmigration from the blood into the tissue.

The peptides or protein according to the invention exhibit the general formula ll

$$\begin{array}{c|c}
R_1 & H & O \\
N - C - C - C - Z_1 - Arg - Z_3 - Z_4 - Z_5 \\
R_2 & H
\end{array}$$

mone, i.e. cortisone, naturally occurring in the body, and 20 wherein R<sub>1</sub> and R<sub>2</sub>, being equal or different, denote hydrogen, a saturated or unsaturated hydrocarbon residue comprising from 1 to 3, in particular up to 10, carbon atoms,

 $Z_1$  denotes a histidine or proline residue,

Arg denotes an arginine,

Z<sub>3</sub> denotes a proline or valine residue,

Z<sub>4</sub> denotes a leucine or valine residue,

Z<sub>5</sub> denotes a protein residue or a peptide residue, in particular comprising

from 2 to 30 amino acids, or an alcohol comprising from 1 to 3, in particular up to 10, carbon atoms, or an organic or inorganic base residue,

as well as the salts thereof, and, f.i., also amides, or mixtures with each other and/or with at least one further substance for therapeutic and/or preventive use in human and/or veterinary medicine, whereby in particular only L-amino acids are provided. Sequences of formula 11 are listed in Table 2.

It was completely surprising that parts of the sequence, peptides or fragments of the fibrinogen exhibit anti-inflammatory effects. Without being bound by such theoretical considerations, said effects might be based on the fact that the fibrin binds to endothelial cells via its neo-N-terminus of the Bbeta-chain and to cells in the bloodstream via the sequence of the Aalpha-chain, thereby leading to the adhesion and transmigration of cells into the tissue. Those bindings exhibit a side reaction in that the formation of fibrin is inhibited. However, said inhibition does not constitute a potential disadvantage to the patient since the blood coagulation is sufficient also in the absence of fibrin if slight injuries occur. Only in case of surgical treatment, it might optionally be suitable to stop such kind of therapy. Other side reactions may substantially be ruled out, since those substances only interact with natural ligands. Furthermore, the natural defence is not affected adversely by the leukocytes in the blood. Thus, the composition of the same, such as granulocytes, lymphocytes and monocytes, remains unaffected so that the natural defence process is maintained and the defence against infections in the blood remains unchanged.

Fibrinogen is produced in the liver and, in this form, is biologically inactive and normally is provided in the blood at concentrations of around 3 g/l. By proteolytic cleavage of the proenzyme prothrombin, thrombin is formed which cleaves off the fibrinopeptides A and B from the fibrinogen. In doing so, fibrinogen is transformed into its biologically active form. Fibrin and fibrin cleavage products are gener-

Thrombin is formed during each activation of the blood coagulation, i.e. with each damage to the tissue, be it of inflammatory, traumatic or degenerative genesis. The formation of fibrin as mediated by thrombin is basically a protective process with the purpose of quickly sealing any defects caused to the vascular system. However, the formation of fibrin is also a pathogenic process. The appearance of a fibrin thrombus as the triggering cause of cardiac infarction is one of the most prominent problems in human medicine.

The role which fibrin plays during the extravastation of inflammatory cells from the bloodstream into the tissue, which, on the one hand, is a desired process of the defence against pathogenic microorganisms or tumour cells occurring in the tissue, but, on the other hand, is a process which, by itself, induces or prolongues damage done to the tissue, has so far not been examined at all or not to a sufficient extent. Fibrin binds to endothelial cells via its neo-Nterminus of Bbeta by means of the sequence to Bbeta and to cells in the bloodstream by means of the sequence Aalpha, 20 thereby leading to the adhesion and transmigration of cells into the tissue.

The peptides or proteins according to the invention may prevent the adhesion of cells from the bloodstream to endothelial cells of the vascular wall and/or their subsequent <sup>25</sup> transmigration from the blood into the tissue.

A peptide or protein according to the invention of the general formula II, wherein  $Z_5$  denotes a peptide residue comprising the following amino acid sequence (SEQ ID NO: 291):

Amp Lys Lys Arg Glu Glu Ala Pro Ser Leu Arg Pro Ala Pro Pro Pro Ile Ser Gly Gly Gly Tyr Arg

and  $Z_1$  denotes a histidine residue, Arg denotes an arginine residue,  $Z_3$  denotes a proline residue,

Z<sub>4</sub> denotes a leucine residue

prevents fibrin fragments from depositing on or adhering to the vascular wall. Thus, it is rendered impossible that inflammatory cells are retained at the endothelial cells of the vascular walls of arteries and veins, and such cells are prevented from remaining at the vascular walls, thus being prevented from infiltrating the tissue any further.

A peptide or protein of the general formula II, wherein  $Z_5$  denotes a peptide residue comprising the following amino acid sequence (SEQ ID NO: 292):

Glu Arg His Gln Ser Ala Cys Lys Asp Ser Asp Trp

Pro Phe Cys Ser Asp Glu Asp Trp Asn Tyr Lys

and  $Z_1$  denotes a proline residue, Arg denotes an arginine residue,  $Z_3$  denotes a valine residue,  $Z_4$  denotes a valine residue

has the effect of preventing the cells of the peripheral blood from adhering to fibrin or finrin fragments, hence prohibiting their migration in the tissue.

The described cleavage products are also known in the literature as peptide Bbeta and peptide Aalpha. Said above 65 mentioned proadhesive and promigratory path is a completely new one for the system of controlling the migration

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of cells from the blood into the tissue. This function of the fibrin may be blocked by peptide Bbeta and also by peptide Aalpha.

Therefore, said peptides according to the invention are suitable as therapeutic agents for humans and animals in order to block the migration of cells from the blood into the tissue. Since fibrin or other fibrinogen products produced by proteolytic cleavage, such as, f.i., fibrinogen cleaved by an urokinase-plasminogen-activator, are generated only to a specific and regionally limited extent, i.e. at sites of inflammation, disturbed coagulation, arterial sclerosis, thrombosis and/or tumour growth, the effect of said therapeutic agent is regionally limited, which means that pathological side effects occurring in other places are not to be expected or only to a limited extent.

Preferable and completely unexpected fields of application for the peptides and/or proteins according to the invention consist in the preparation of pharmaceutical compositions for the therapy or prevention of local and/or generalized inflammations in the body in case of infectious genesis, based upon an auto-immune reaction, based upon a rheumatic disease, based upon a disorder in the immune system, based upon a genetic disease, for the prevention and/or therapy of the rejection occurring after organ transplants, of arterial sclerosis, of a reperfusion trauma, based upon arteriosclerotic and/or thrombotic diseases and an increased fibrin deposition. Such a peptide, in particular Bbeta, is also excellently suitable for the preparation of a pharmaceutical composition which accomplishes the transportation of a further drug substance to human or animal endothelial cells. In doing so, the drug substance to be transported is coupled to the peptide at one end and then, via VE-cadherin, deposits on a free spot of the vascular wall, i.e. on an endothelial cell.

In the following, the invention is explained in further detail by way of examples.

## EXAMPLE 1

Preparation of the Fibrinogen Cleavage Products:

Non-polymerizing degradation products of fibrinogen were obtained by means of a decomposition involving cyanogen bromide according to Blombäck et al. (Nature 1968, 218; 130-134). The fibrinogen thus degraded largely consists of a 63 kD fragment, i.e. the N-terminal disulfide knot, NDSK, and comprises Aalpha-chain 1-51, Bbeta-chain 1-118 and gamma-chain 1-78. In order to obtain NDSK-II (NDSK minus fibrinopeptides A and B), the N-terminal amino acids of the Aalpha- and Bbeta-chains were cleaved off with thrombin (20 units/1 µg NDSK) in three hours at room temperature and subsequently were treated with diisopropylfhuorophosphate in order to block thrombin activity. The NDSK-II thus obtained consisted of Aalpha-chain 17-51, Bbeta-chain 15-118 and gamma-chain 1-78.

In order to obtain NDSK-uPA, 500 µg of NDSK was treated with 200 units of urokinase-plasminogen-activator (uPA) of Messrs. Technoclone, Vienna, Austria, for one hour at 37° C. The reaction was stopped with 5 mM phenylm-ethylsulfonyl fluoride. The NDSK-uPA thus obtained is a NDSK and has no fibrinopeptide B.

As a negative control, a second fraction was obtained from the fibrinogen cleavage products referred to as FCB-2 according to Nieuwenhuizen et al. (Biochem Biophys Acta 1983, 715; 531-533), which cleavage products were produced by being treated with cyanogen bromide. FCB-2 is a protein having a size of 43 kD and consists of Aalpha-chain

148-208, Bbeta-chain 191-305 and gamma-chain 95-265. For control purposes, thrombin and diisopropylfluorophosphate were added to said protein. That, however, did not result in any change to the protein (in the following, referred to as FCB-2-thr).

For the purpose of further negative controls, culture medium (RPMI of Messrs. Life techn. Inc., Paisky, UK) was treated with thrombin as above and, subsequently, was inactivated (RPMI-thr) or was treated with uPA as above and was inactivated (RPMI-uPA).

### **EXAMPLE 2**

Peptide Aalpha (SEQ 1D NO: 293) corresponds to amino acids I to 28 of the alpha chain of the fibrin and is identical to amino acids 17 to 45 of the Aalpha chain of the fibrinogen:

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e) 100 µg of randomized Aalpha

f) 100 μg of randomized Bbeta

Twenty-four hours later, the human skin was removed and the number of inflammatory sites, expressed in cells per 0.3 mm<sup>2</sup>, was evaluated and the mean value was determined with a standard deviation.

For a: 22+/-2.8

10 for b: 9+/-2.I

for c: 4+/-1.1

for d: 6+/-1.1

for e: 5+/-1.2

for f: 7+/-1.3

Gly Pro Arg Val Val Glu Arg His Gln Ser Ala Cys Lys
Asp Ser Asp Trp Pro Phe Cys Ser Asp Glu Asp Trp Asn
Tyr Lys

Peptide Bbeta (SEQ 1D NO: 294) corresponds to amino 25 acids I to 28 of the beta chain of the fibrin and is identical to amino acids 15 to 43 of the Bbeta chain of the fibrinogen, which exhibits the following sequence:

That allows the conclusion that NDSK-II causes inflammations, and hence said protein was used as a pathogenic substance. The other compounds per se do not exhibit any significant increase in the amount of inflammatory cells.

Gly His Arg Pro Leu Asp Lys Lys Arg Glu Glu Ala Pro Ser Leu Arg Pro Ala Pro Pro Pro Ile Ser Gly Gly Tyr Arg

By applying a fluorenylmethyloxycarbonyl (FMOC)-protective group strategy according to Carpino L. A. and Han. G Y, J. Amer. Chem. Soc. 1981; 37; 3404-3409, both peptides were synthesized by means of a solid-phase peptide 40 synthesis according to Merrifield R. B., J. Amer. Chem. Soc. 1963; 85, 2149-2154, using a multiple peptide synthesizer. The crude peptides were purified by preparative reversed-phase HPLC via a Nucleosil 100-10, C18-column according to Engelhart H. and Müller H. Chromatography 1984 19:77 as well as Henschen A., Hupe K. P. and Lottspeich F. High Performance Liquid Chromatography VCH 1985. As control peptides, peptides of the same length but comprising a randomized amino acid sequence were used.

### EXAMPLE 3

### **HU-SCID Mouse-Model:**

Human skin was transplanted onto the backs of SCID mice, and two weeks later human lymphocytes were injected into the peritoneum. The proceedings were according to Petzelbauer et al. (J. Invest. Dermatol. 1996, 107; 576-581). Then, fifteen mice thus prepared were injected in their tail veins with the following:

- a) 100 µg of human NDSK-II
- b) 100 μg of human FCB-2
- c) I00 µg of peptide Aalpha
- d) 100 µg of peptide Bbeta

# COMPARATIVE EXAMPLE 4

Fifteen mice according to Example 3 were injected in their tail veins with

100 μg of human NDSK-II and

100 μg of randomized peptide Aalpha.

Further proceedings were according to Example 3. Per 0.3 mm<sup>2</sup>, 23+/-3.5 inflammatory sites could be determined.

### COMPARATIVE EXAMPLE 5

Fifteen mice according to Example 3 were injected in their tail veins with

 $100~\mu g$  of human NDSK-II according to Example I and  $100~\mu g$  of randomized peptide Bbeta.

Further proceedings were according to Example 3. Per 0.3 mm<sup>2</sup>, 24+/-2 inflammatory sites could be determined.

# EXAMPLE 6

Fifteen mice according to Example 3 were injected with

100 µg of human NDSK-II and

65 I00 μg of synthesized peptide Aalpha.

Further proceedings were according to Example 3. Per 0.3 mm<sup>2</sup>, 21+/-2.2 inflammatory sites could be determined.

# 7 EXAMPLE 7

Fifteen mice according to Example 3 were injected in their tail veins with

100 µg of human NDSK-11 and

100 μg of synthesized peptide Bbeta.

Further proceedings were according to Example 3. Per 0.3 mm<sup>2</sup>, 14+/-2 inflammatory sites could be determined.

Examples 4 to 7 show that peptide Bbeta blocks lymphocytic inflammation.

### COMPARATIVE EXAMPLE 8

Endothelial cells from human umbilical veins (HUVEC) were marked with a red fluorescent dye (Cell Tracker Orange, 1 µl/ml, Molecular Probes, Eugene, Oreg.) and were dispersed on a collagen matrix (Collaborative Biomedical Products, Bedford, Mass.). Upon confluence of the endothelial cells, peripheral mononuclear blood cells (PBMC) (10<sup>5</sup> cells per 25 mm²) marked with a green fluorescent dye (Cell Tracker Green, 1 µl/ml, Molecular Probes of Messrs. Eugene, Origon) were superimposed. Thereafter, the cells were incubated at 37° C. for twelve hours.

Adhering cells that had transmigrated into the gel were photographed with a laser-scan microscope, were converted into pixels and were evaluated by means of an "NIH image" according to Gröger et al. (J. Immunol. Method 1999; 222: 101-109).

It was feasible to determine the number of adherent cells per 0.1 mm<sup>2</sup> such as mentioned under "adhesion". It was 35 feasible to determine the number of migrated cells per 0.04 mm<sup>3</sup> such as mentioned under "migration". The mean value of three times three trials was evaluated together with the standard deviation.

			adhesion	migration	
a)	RPMI-uPA	0.1 µg/ml	40 +/- 4	4 +/- 3	_
		1.0 μg/ml	38 +/- 2	5 +/- 2	
		10.0 μg/ml	32 +/- 4	5 +/- 1	
b)	NDSK	0.1 μg/ml	31 +/- 18	6 +/- 3	
		1.0 μg/ml	35 +/- 18	5 +/- 2	
		10.0 μg/ml	36 +/- 24	6 +/- 3	
c)	NDSK-II	0.1 μg/ml	55 +/ 21	12 +/- 5	
		1.0 μg/ml	67 +/- 31	19 +/- 12	
		10.0 μg/ml	65 +/- 31	19 +/- 10	
d)	NDSK-uPA	0.1 μg/ml	58 +/- 3	10 +/- 2	
		1.0 μg/ml	60 +/- 3.5	14 +/- 3	
		10.0 μg/ml	65 +/- 3	18 +/- 1.5	
e)	FCB2	0.1 μg/ml	30 +/- 26	6 +/- 4	
		1.0 μg/ml	10 +/- 10	3 +/- 2	
		10.0 μg/ml	21 +/- 7	5 +/- 4	
f)	FCB-2-thr	0.1 μg/ml	20 +/- 12	6 +/- 5	
		1.0 μg/ml	23 +/- 13	7 +/- 5	
		10.0 μg/ml	26 +/- 11	4 +/- 2	
g)	RPMI-thr	0.1 μ <b>g/ml</b>	29 +/- 15	4 +/- 5	
		1.0 µg/ml	26 +/- 14	5 +/- 5	
		10.0 μg/ml	41 +/- 20	5 +/- 4	

That allows the conclusion that NDSK-II results in significant migrations of peripheral blood-monocellular cells (PBMC) to a greater extent than NDSK-uPA and hence 65 exhibits pathogenic activity. None of the controls a), b), e), f) and g) resulted in any significant migration.

# **8** EXAMPLE 9

100 μg of NDSK-11 and Bbeta or Bbeta randomized were added to the collagen matrix according to Example 8 comprising the suspension of PBMC, and further proceedings were in accordance with Example 8.

		adhesion	migration
a) b) c) d) e) f) g)	no addition of NDSK-II only 100 µg of NDSK-II 10 µg of Bbeta + NDSK-II 100 µg of Bbeta + NDSK-II 1000 µg of Bbeta + NDSK-II 1000 µg of Bbeta + NDSK-II 1000 µg of Bbeta randomized + NDSK-II 1000 µg of Bbeta randomized + NDSK-II 1000 µg of Bbeta randomized + NDSK-II	38 +/- 15 73 +/- 29 63 +/- 33 47 +/- 34 52 +/- 27 77 +/- 33 86 +/- 35 78 +/- 31	6 +/- 4 16 +/- 7 7 +/- 4 5 +/- 4 10 +/- 6 16 +/- 6 15 +/- 6 13 +/- 8

As can be gathered from those test results, peptide Bbeta blocks inflammations.

### **EXAMPLE 10**

100 μg of NDSK-II and Aalpha or Aalpha randomized were added to the collagen matrix according to Example 8 comprising the suspension of PBMC, and further proceedings were in accordance with Example 8.

)		adhesion	migration
a)	no addition of NDSK-II	42 +/- 6	10 +/- 1
ь)	only NDSK-II	96 +/- 11	24 +/- 3
c)	10 μg of Aalpha + NDSK-II	69 +/- 12	21 +/- 4
d)	100 μg of Aalpha + NDSK-II	73 +/- 13	15 +/- 6
6 e)	1000 µg of Aalpha + NDSK-II	70 +/- 6	13 +/- 5
f)	10 µg of Aalpha randomized + NDSK-II	70 +/- 6	25 +/- 2
g)	100 µg of Aalpha randomized + NDSK-II	65 +/- 16	24 +/- 3
h)	1000 μg of Aalpha randomized + NDSK-II	70 +/- 12	26 +/- 3

As can be gathered from the test results, peptide Aalpha blocks the migration of PBMC only partially.

### EXAMPLE 11

Since PBMC substantially consists of a mixture of lymphocytes and monocytes, pure lymphocytes instead of PBMC (as in Examples 8-10) were used in Example 11.

100 µg of NDSK-uPA or 100 µg of NDSK-II, respectively, and Aalpha or Bbeta, respectively, were added to the collagen matrix according to Example 8 comprising endothelial cells and lymphocytes.

		adhesion	migration
a)	no addition	68 +/- 8	16 +/- 3
b)	NDSK-uPA	143 +/- 11	53 +/- 5
c)	NDSK-II	119 +/- 11	43 +/- 4
d)	only 100 µg of Bbeta	58 +/- 18	14 +/- 1
e)	NDSK-uPA + 100 µg of Bbeta	74 +/- 8	19 +/- 2
f)	NDSK-II + 100 µg of Bbeta	74 +/- 8	17 +/~ 3
g)	only 100 µg of Aalpha	77 +/- 4	18 +/- 1
1)	NDSK-uPA + 100 µg of Aalpha	131 +/- 4	40 +/- 3
)	NDSK-II + 100 µg of Aaipha	131 +/- 4	44 +/- 4
)	only 100 µg of Bbeta randomized	75 +/- 5	19 +/- 1
<b>(</b> )	NDSK-uPA + 100 µg of Bbeta randomized	134 +/- 13	46 +/- 4
l)	NDSK-II + 100 µg of Bbeta randomized	120 +/- 12	42 +/- 4

Those test results show

- that both NDSK-II and NDSK-uPA promote lymphocytic inflammation.
- 2) that peptide Bbeta completely blocks the lymphocytic adhesion and migration induced by NDSK-II and NDSKuPA, whereas peptide Aalpha exhibits no blocking activity, which suggests that the free alpha-chain is not required for inducing the adhesion and migration of the lymphocytes.

## EXAMPLE 12

The proceedings were in accordance with Example 11, except for pure monocytes being used instead of lymphocytes. 100 µg of NDSK-uPA or 100 µg of NDSK-II, respectively, was added to peptide Aalpha, randomized Aalpha, Bbeta or randomized Bbeta.

		adhesion	migration
a)	no addition	43 +/- 8	7 +/- 1
b)	NDSK-uPA	48 +/- 10	10 +/- 2
2)	NDSK-II	90 +/- 11	19 +/- 6
(t	100 μg of Bbeta	59 +/- 7	5 +/- 1
e)	NDSK-uPA + 100 μg of Bbeta	61 +/- 11	8 +/- 3
D)	NDSK-11 + 100 µg of Bbeta	70 +/- 7	7 +/- 5
g)	100 µg of Bbeta randomized	40 +/- 7	6 +/- 1
n)	NDSK-uPA + 100 µg of Bbeta randomized	45 +/- 5	8 +/- 3
g)	NDSK-II + 100 µg of Bbeta randomized	92 +/- 10	20 +/- 7
)	100 µg of Aalpha	59 +/- 6	5 +/- 1
(2	NDSK-uPA + 100 µg of Aalpha	62 +/- 4	8 +/- 5
)	NDSK-I1 + 100 µg of Aalpha	68 +/- 10	9 +/- 6
n)	100 µg of Aalpha randomized	58 +/- 7	6 +/- 1
1)	NDSK-uPA + 100 μg of Aalpha randomized	50 +/- 10	10 +/- 4
)	NDSK-11 + 100 μg of Aalpha randomized	108 +/- 8	21 +/- 5

Those test results show that only NDSK-II and not NDSK-uPA promotes the migration of monocytes, which means that both the alpha-chain and the beta-chain have to exhibit a free N-terminal end and block the migration of the monocytes.

## **EXAMPLE 13**

The proceedings were in accordance with Example 11, 45 with pure lymphocytes being used. 100 µg of NDSK-uPA or 100 µg of NDSK-ll, respectively, was added to the short peptide salts derived from Aalpha Gly Pro Arg (Pro)—NH<sub>2</sub> acetate (Aalpha derivative) or derived from Bbeta Gly His Arg Pro-OH acetate (Bbeta derivative).

		adhesion	migration
a)	no addition	60 +/- 8	14 +/- 1
b)	NDSK-uPA	149 +/- 12	57 +/- 5
c)	NDSK-II	121 +/- 11	48 +/- 7
d)	only 100 μg of Bbeta derivative	58 +/- 10	12 +/- 9
e)	NDSK-uPA + 100 µg of Bbeta derivative	70 +/- 8	16 +/- 3
f)	NDSK-11 + 100 µg of Bbeta derivative	69 +/- 7	14 +/- 5
g)	only 100 μg of Aalpha derivative	77 +/- 4	18 +/- 1
h)	NDSK-uPA + 100 µg of Aalpha derivative	134 +/- 4	48 +/- 5
i)	NDSK-II + 100 µg of Aalpha derivative	131 +/- 7	49 +/- 6
j)	only 100 µg of Bbeta derivative randomized	70 +/- 5	14 +/- 7
k)	NDSK-uPA + 100 µg of Bbeta derivative		
1)	randomized NDSK-II + 100 µg of Bbeta derivative	130 +/- 12	49 +/- 6
	randomized	120 +/- 10	55 +/- 8

Said experiment allows the conclusion that, if lymphocytic migration is inhibited, those short peptides, added continuously in an appropriate manner, exhibit the same activity as do the long peptides.

### **EXAMPLE 14**

The proceedings were in accordance with Example 12, with pure monocytes being used. 100 mg of NDSK-uPA or 100 μg of NDSK-II, respectively, was added to the short peptide salts Aalpha Gly Pro Arg (Pro)—NH<sub>2</sub> acetate (Aalpha derivative) or Bbeta Gly His Arg Pro-OH acetate (Bbeta derivative).

			adhesion	migration
	a)	no addition	40 +/- 8	5 +/- 1
	b)	NDSK-uPA	54 +/- 9	7 +/- 2
20	c)	NDSK-II	85 +/- 11	22 +/- 6
20	d)	100 μg of Bbeta derivative	52 +/- 7	6 +/- 1
	e)	NDSK-uPA + 100 µg of Bbeta derivative	61 +/- 11	8 +/- 3
	f)	NDSK-I1 + 100 µg of Bbeta derivative	68 +/- 7	8 +/- 4
	g)	100 μg of Bbeta derivative randomized	40 +/- 7	6 +/- 1
	h)	NDSK-uPA + 100 µg of Bbeta derivative	44 +/- 6	8 +/- 2
	_	randomized		
25	i)	NDSK-11 + 100 μg of Bbeta derivative	92 +/- 10	23 +/- 7
	j)	100 µg of Aalpha derivative	50 +/- 5	4 +/- 4
	k)	NDSK-uPA + 100 µg of Aalpha derivative	60 +/- 5	7 +/- 6
	(1	NDSK-1I + 100 µg of Aalpha derivative	64 +/- 11	8 +/- 2
	m)	100 μg of Aalpha derivative randomized	54 +/- 10	6 +/- 3
30	n)	NDSK-uPA + 100 µg of Aalpha derivative	50 +/- 10	10 +/- 4
		randomized		
	0)	NDSK-II + 100 µg of Aalpha derivative	99 +/- 8	21 +/- 7
	,	randomized		
		randomized		

Said experiment allows the conclusion that, if monocytic migration is inhibited, those short peptides, added continuously in an appropriate manner, exhibit the same activity as do the long peptides.

### **EXAMPLE 15**

The tests were carried out on male wistar rats weighing between 220 g and 280 g. The rats were given standard food and water. For carrying out the test, the rats were anaesthetized and artifically respirated with a frequency of 70 pulses per minute, whereby from 8 ml to 10 ml per kilogram of a gas containing 30% by volume of oxygen and having an overpressure of from 1 mm to 2 mm mercury was emitted. The cardiac artery on the right hand side was equipped with 50 a measuring cannula, and the blood pressure in the artery as well as the heartbeats were determined. The pressure rate was determined as a product of the blood pressure in the artery and of the heartbeat rate with the dimension mm mercury/minute/10<sup>3</sup>. The vein on the right hand side was 55 equipped with a measuring cannula for doping the test substances. After carrying out the surgical treatment, 2 ml of rat blood was supplied to the heart. Thirty minutes later, the cardiac artery on the left hand side was occluded. Another twenty-five minutes later, the occlusion was released in 60 order to resupply the ischaemic area with blood. At that point of time, 800 μg/kg of peptide Bbeta or peptide Bbeta randomized, respectively, was intravenously administered to half of the animals, and then two hours were allowed to pass.

In order to distinguish between damaged and undamaged cardiac tissue, the cardiac artery on the left hand side was then supplied with evans blue dye at a concentration of 2% by weight. Thereupon, the removed heart was dissected by

five horizontal cuts, the right hand wall of the vein was removed and the sections were treated with triphenyltetratolchloride (1% by weight) for twenty minutes at 37° C. so as to be able to distinguish between normal tissue and infarct tissue. The sections were evaluated by computer-sustained 5 planimetry.

Because of the vascular occlusion, 62.5% of the cardiac muscle in the hearts of the reference rats was threatened, as opposed to 60% in the hearts of the test rats. In the hearts of the reference rats, 46% of the endangered tissue was dead, 10 as opposed to 29% in the hearts of the test rats. That corresponds to a 37% reduction of dead tissue (p<0.05).

The substances according to the invention as well as the use of the substances according to the invention for preparing a pharmaceutical composition are of special signifi- 15 cance:

For a pharmaceutical composition used in the therapy of diseases caused by the tissue-damaging effect of autoreactive lymphocytes.

Among those are diseases fitting into the sphere of 20 autoimmunity, such as collagenoses, rheumatic diseases, psoriasis and post-/parainfectious diseases and diseases caused by a graft versus host reaction. A healing effect occurs, since said pharmaceutical composition blocks the migration of lymphocytes into the tissue. Thus, the lymphocytes remain in the bloodstream and are incapable of producing an autoreactive tissue-damaging effect.

A healing effect occurs with a drug for the therapy and/or prevention of the rejection occurring after organ transplants, since said drug prevents the migration of lymphocytes from 30 the bloodstream into the foreign organ and hence the foreign organ cannot be destroyed by autoreactive lymphocytes.

A healing effect occurs with a drug for the therapy and/or prevention of arterial sclerosis after organ transplants, since said drug prohibits the migration of lymphocytes and monocytes into the vascular wall and hence prevents the activation of the cells of the vascular wall. In doing so, the occurrence of arterial sclerosis following organ transplants is minimized or prevented.

A healing effect occurs with a drug for the therapy and/or 40 prevention of a reperfusion trauma following a surgically or pharmaceutically induced restoration of the blood flow such as, f.i. after cardiac infarction, apoplectic stroke, after vascular surgery, bypass surgery and organ transplants, since said drug inhibits the migration of lymphocytes and mono- 45 cytes into the vascular wall. The reperfusion trauma is caused by oxygen deficiency/acidosis occurring in the cells of the vessel during the restoration of the blood flow and leads to their activation. Thereby, lymphocytes and monocytes adhere to the vascular wall and migrate into the same. 50 The fact that lymphocytes and monocytes are prevented from adhering to and migrating into the vascular wall brings about a decrease in the hypoxia/acidosis-induced damage, without any permanent vascular damage being caused by the subsequent inflammatory reaction.

A healing effect occurs with a drug for the therapy and/or prevention of arterial sclerosis following metabolic diseases or ageing processes, since said drug inhibits the migration of lymphocytes and monocytes into the vascular wall and hence inhibits the progredience of the arteriosclerotic plaque 60 resulting therefrom.

The pharmaceutical composition according to the invention may also be used for transporting a further drug substance. The pharmaceutical composition according to the invention specifically binds a surface molecule to endothelial cells. Thus, drug substances coupled thereto may be contacted with endothelial cells at high concentrations,

without them being able to trigger side reactions in other places. The use of substances inhibiting cell division may be mentioned as an example, which substances may exhibit an antiangiogenetic effect after having been adducted specifically to endothelial cells. In that case, tumour patients experience a healing effect, since the growth of the tumour is blocked by preventing the proliferation of endothelial cells and hence by avoiding neoangiogenesis.

12

TABLE 1

Peptides of Formula 1; Glv-His/Pro-Arg-Xaa2-Xaa20						
	SEQ					
SECULENCE	ID					
SEQUENCE	NO					
Gly His Arg Gly Pro Arg	1 2					
Gly His Arg Xaa	3					
Gly Pro Arg Xaa	4					
Gly His Arg Xaa Xaa	5					
Gly Pro Arg Xaa Xaa Gly His Arg Xaa Xaa Xaa	6 7					
Gly Pro Arg Xaa Xaa Xaa	8					
Gly His Arg Xaa Xaa Xaa Xaa	9					
Gly Pro Arg Xaa Xaa Xaa Xaa	10					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Clas Baa Ara Xaa Xaa Xaa	11					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa	12 13					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa	14					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa	15					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa	16					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	17					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Cly His Are Yee Xee Xee Xee Xee Xee Xee Xee Xee Xe	18					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	19 20					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	21					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	22					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	23					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	24					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	25					
Xaa Giy Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	26					
Xaa Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	27					
Xaa Xaa						
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	28					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	29					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	30					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	31					
Xaa	32					
Xaa	295					
Xaa Xaa Xaa Xaa						
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	296					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	33					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	34					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	35					
Xaa Xaa Xaa Xaa Xaa Xaa Xaa Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	36					
Xaa	37					
Xaa Xaa Xaa Xaa Xaa Xaa Xaa						
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	38					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	39					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	40					
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	41					
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	42					

TABLE 1-continued

14

Peptides of Formula I: Gly-His/Pro-Arg-Xaa2-Xaa29		_	Peptides of Formula I: Gly-His/Pro-Arg-Xaa <sub>2</sub> -Xaa <sub>29</sub>	
SEQUENCE	SEQ ID NO	5	SEQUENCE	SEQ ID NO
Xaa	43	10	Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	52
Xaa	44		Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	53
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	45 46	15	Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	54
Xaa	47	20	Xaa Xaa Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	55
Xaa	48	20	Xaa Xaa Xaa Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	56
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	49	25	Xaa	57
Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	50	23	Xaa Xaa Xaa Xaa Gly Pro Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	58
Gly His Arg Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	51		Xaa	

TABLE 2

EQUENCE	SEQ ID NO
ly His Arg Pro Leu Xaa Xaa	59
ly Pro Arg Pro Leu Xaa Xaa	60
ly His Arg Val Leu Xaa Xaa	61
ly Pro Arg Val Leu Xaa Xaa	62
ly His Arg Pro Val Xaa Xaa	63
ly Pro Arg Pro Val Xaa Xaa	64
ly His Arg Val Val Xaa Xaa	65
ly Pro Arg Val Val Xaa Xaa	66
ly His Arg Pro Leu Xaa Xaa Xaa	67
ly Pro Arg Pro Leu Xaa Xaa Xaa	68
ly His Arg Val Leu Xaa Xaa Xaa	69
ly Pro Arg Val Leu Xaa Xaa Xaa	70
ly His Arg Pro Val Xaa Xaa Xaa	71
ly Pro Arg Pro Val Xaa Xaa Xaa	72

Pe	ptide	s of	For	mula	II:	Gly	-His	/Pro	-Arc	-Pro	/ <u>Val-</u>	Leu/Va	l-Xaa <sub>2</sub> -X6	18 <sub>30</sub>		
SEQUENCE														SEQ	ID	NO
Gly His Arg	Val	Val	Xaa	Xaa	Xaa										73	
Gly Pro Arg	Val	Val	Xaa	Xaa	Xaa										74	
Gly His Arg	Pro	Leu	Xaa	Xaa	Xaa	Xaa									75	
Gly Pro Arg	Pro	Leu	Xaa	Xaa	Xaa	Xaa									76	
Gly His Arg	Val	Leu	Xaa	Xaa	Xaa	Xaa									77	
Gly Pro Arg	Val	Leu	Xaa	Xaa	Xaa	Xaa									78	
Gly His Arg	Pro	Val	Xaa	Xaa	Xaa	Xaa									79	
Gly Pro Arg	Pro	Val	Xaa	Xaa	Xaa	Xaa									80	
Gly His Arg	Val	Val	Xaa	Xaa	Xaa	Xaa									81	
Gly Pro Arg	Val	Val	Xaa	Xaa	Xaa	Xaa									82	
Gly His Arg	Pro	Leu	Xaa	Xaa	Xaa	Xaa	Xaa								83	
Gly Pro Arg	Pro	Leu	Xaa	Xaa	Xaa	Xaa	Xaa								84	
Gly His Arg	Val	Leu	Xaa	Xaa	Xaa	Xaa	Xaa								85	
Gly Pro Arg	Val	Leu	Xaa	Xaa	Xaa	Xaa	Xaa								86	
Gly His Arg	Pro	Val	Xaa	Xaa	Xaa	Xaa	Xaa								87	
Gly Pro Arg	Pro	Val	Xaa	Xaa	Xaa	Xaa	Xaa								88	
Gly His Arg	Val	Val	Xaa	Xaa	Xaa	Xaa	Xaa								89	
Gly Pro Arg	Val	Val	Xaa	Xaa	Xaa	Xaa	Xaa								90	
Gly His Arg	Pro	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa							91	
Gly Pro Arg															92	
Gly His Arg															93	
Gly Pro Arg															94	
Gly His Arc															95	
Gly Pro Arg															96	
Gly His Arg															97	
Gly Pro Arg									V						98	
Gly Bro Are															99 100	
Gly His Arg															101	
Gly Pro Arc															102	
Gly His Arc															103	
Gly Pro Arc															104	
Gly His Arg															105	
Gly Pro Arg															106	
Gly His Arg	J Pro	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa					107	
Gly Pro Arg	, Pro	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa					108	
Gly His Arg	Val	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa					109	

Peptides of Formula II: Gly-His/Pro-Arq-Pro/Val-Leu/Val-Xaa2-X	aa <sub>30</sub>
SEQUENCE	SEQ ID NO
Gly Pro Arg Val Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	110
Gly His Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	111
Gly Pro Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	112
Gly His Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	113
Gly Pro Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	114
Gly His Arg Pro Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	115
Gly Pro Arg Pro Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	116
Gly His Arg Val Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	117
Gly Pro Arg Val Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	118
Gly His Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	119
Gly Pro Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	120
Gly His Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	121
Gly Pro Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa	122
Gly His Arg Pro Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	123
Gly Pro Arg Pro Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	124
Gly His Arg Val Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	125
Gly Pro Arg Val Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	126
Gly His Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	127
Gly Pro Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	128
Gly His Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	129
Gly Pro Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	130
Gly His Arg Pro Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	131
Gly Pro Arg Pro Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	132
Gly His Arg Val Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	133
Gly Pro Arg Val Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	134
Gly His Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	135
Gly Pro Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	136
Gly His Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	137
Gly Pro Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	138
Gly His Arg Pro Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	139
Gly Pro Arg Pro Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	140
Gly His Arg Val Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	141
Gly Pro Arg Val Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	142
Gly His Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	143
Gly Pro Arg Pro Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	144
Gly His Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	145
Gly Pro Arg Val Val Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa X	146
Gly His Arg Pro Leu Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xaa Xa	147

Peptides of Formula II: Gly-His/Pro-Arg-Pro/Val-Leu/Val-Xaa2-Xaa30																			
SEQ	UENC	E					_											SEQ I	ON C
			_												_				
-		,															Хаа	14	
_		-															Xaa	14	
_																	Xaa	15	
-		-															Xaa 	15	
-		-															Xaa	15:	
-																	Xaa 	15:	
-		_															Xaa	15	
Xaa		Arg	Pro	Leu	хаа	хаа	Xaa	хаа	Xaa	15!	,								
Gly Xaa	Pro	Arg	Pro	Leu	Xaa	150	5												
Gly Xaa	His	Arg	Val	Leu	Xaa	15	7												
Gly Xaa	Pro	Arg	Val	Leu	Xaa	Хаа	Xaa	Xaa	Xaa	Xaa	Xaa	150	3						
Gly Xaa	His	Arg	Pro	Val	Xaa	159	)												
Gly Xaa	Pro	Arg	Pro	Val	Xaa	160	)												
Gly Xaa	His	Arg	Val	Val	Xaa	16	L												
Gly Xaa	Pro	Arg	Val	Val	Xaa	162	?												
	His Xaa	Arg	Pro	Leu	Xaa	163	3												
	Pro Xaa	Arg	Pro	Leu	Xaa	164	ļ												
	His Xaa	Arg	Val	Leu	Xaa	165	;												
	Pro Xaa	Arg	Val	Leu	Xaa	166	i												
Gly Xaa	His Xaa	Arg	Pro	Val	Xaa	167	r												
	Pro Xaa	Arg	Pro	Val	Xaa	168	I												
Gly Xaa	His Xaa	Arg	Val	Val	Xaa	169	ı												
Gly Xaa	Pro Xaa	Arg	Val	Val	Xaa	170													
Gly Xaa	His Xaa	Arg Xaa	Pro	Leu	Xaa	171													
Gly Xaa	Pro Xaa	Arg Xaa	Pro	Leu	Xaa	172													
Gly Xaa	His Xaa	Arg	Val	Leu	Xaa	173													
Gly Xaa	Pro Xaa	Arg Xaa	Val	Leu	Xaa	174													

_	Peptides of Formula II: Gly-His/Pro-Arg-Pro/Val-Leu/Val-Xaa2-Xaa30																		
SEQUENCE SEQ ID NO														ID NO					
	His Xaa		Pro	Val	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		175
	Pro Xaa		Pro	Val	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		176
_	His Xaa	_		Val	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		177
	Pro Xaa		Val	Val	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		178
	His Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		179
-	Pro Xaa	_		Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		180
-	His Xaa	_			Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		181
	Pro Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		182
	Hia Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		183
	Pro Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		184
	His Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		185
	Pro Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		186
	His Xaa					Xaa	Xaa	Xaa	Xaa		187								
	Pro Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		188
	His Xaa					Xaa	Xaa	Xaa	Xaa		189								
	Pro Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		190
-	His Xaa	-				Xaa	Xaa	Xaa	Xaa		191								
	Pro Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Хаа	Xaa	Xaa	Xaa	Xaa	Xaa		192
	His Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		193
	Pro Xaa				Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa		194
	His Xaa				Xaa Xaa	Xaa	Xaa	Xaa	Xaa		195								
	Pro Xaa				Xaa Xaa	Xaa	Xaa	Xaa	Xaa		196								
	His Xaa				Xaa Xaa	Xaa	Xaa	Xaa	Xaa		197								
Xaa	Xaa	Xaa	Xaa	Xaa															198
Gly Xaa	His Xaa	Arg Xaa	Pro Xaa	Val Xaa	Xaa Xaa	Xaa	Xaa	Xaa	Xaa		199								

Peptides of Formula II: Gly-His/Pro-Arq-Pro/Val-Leu/Val-Xaa,-Xaa10														_					
SEQ	UENC	E																SEQ ID	NO
	Pro Xaa						Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	200	_
	His Xaa						Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	201	
	Pro Xaa						Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	202	
Gly		Arg	Pro	Leu	Xaa	Xaa		Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	203	
Gly		Arg	Pro	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	204	
Gly	His	Arg	Va 1	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	205	
Gly		Arg	Val	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	206	
	Xaa His							Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	207	
	Xaa Pro							Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	208	
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa											Xaa	209	
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa													
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa											Xaa	210	
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa										Xaa	211	
	Pro Xaa								Xaa	212									
	His Xaa								Xaa	213									
	Pro Xaa								Xaa	214									
	His Xaa								Xaa	215									
	Pro Xaa								Xaa	216									
Gly Xaa	His Xaa	Arg Xaa	Val Xaa	Val Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	217	
Gly Xaa	Pro Xaa	Arg Xaa	Val Xaa	Val Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	218	
Gly Xaa	His Xaa	Arg Xaa	Pro Xaa	Leu Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa	219									
Gly Xaa	Pro Xaa	Arg Xaa	Pro Xaa	Leu Xaa	Xaa Xa <i>a</i>	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa	220									
Gly Xaa	His Xaa	Arg Xaa	Val Xaa	Leu Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa	221									
Gly Xaa	Pro Xaa	Arg Xaa	Val Xaa	Leu Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa	222									
Gly	His Xaa	Arg	Pro	Val	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	223	
Gly Xaa	Pro Xaa	Arg Xaa	Pro Xaa	Val Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa Xaa	Xaa	224									

	_	Pe	otid	e <u>s o</u>	f Fo	rmula	a II	: Gl	y-Hi	s/Pr	o-Ar	q-Pr	o/Va	l-Le	u/Va	l-Xa	a <sub>z</sub> -Xaa	a <sub>30</sub>		
SEQ	JENCI	£																SEQ	ID	N
						Xaa Xaa			Xaa	2	25									
-		_				Xaa Xaa				Xaa	2	26								
-		-				Xaa Xaa					Xaa	2	27							
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						Xaa Xaa				Xaa	2	33								
Gly	Pro	Arg	Val	Val	Xaa		Xaa	2	34											
Gly	His	Arg	Pro	Leu	Xaa		Xaa	Xaa	Xaa		Xaa	2	35							
Gly	Pro	Arg	Pro	Leu	Xaa		Xaa	2	36											
Gly	His	Arg	Val	Leu	Xaa		Xaa	2	37											
Gly	Pro	Arg	Val	Leu	Xaa		Xaa	2	38											
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Gly	Pro	Arg	Pro	Val	Xaa		Xaa	2	40											
Gly	His	Arg	Val	Val	Xaa		Xaa	2	41											
Gly	Pro	Arg	Val	Val	Xaa		Xaa	2	42											
Gly	His	Arg	Pro	Leu	Xaa		Xaa	Xaa	Xaa	Xaa		Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	2	4.3	
Gly	Pro	Arg	Pro	Leu	Xaa		Xaa	2	44											
						Xaa Xaa						Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	2	45	
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa					Xaa		2	46	
Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa					Xaa			47	
Kaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa					Xaa			48	
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\aa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	vag	vaq	AG G	naa	naa	vaq	2	49	

TABLE 2-continued

	_											nue	_		_				
Peptides of Formula II: Gly-His/Pro-Arq-Pro/Val-Leu/Val-Xaa,-Xaa,																			
SEQU	JENCE						_		_									SEQ I	D NO
Gly Xaa	Pro Xaa	Arg Xaa	Val Xaa	Val Xaa	Xaa Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	25	50						
Gly Xaa	His Xaa	Arg Xaa	Pro Xaa	Leu Xaa	Xaa Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	25	51							
	Pro Xaa												Xaa	Xaa	Xaa	Xaa	Xaa	25	52
	His Xaa												Xaa	Xaa	Xaa	Xaa	Xaa	25	3 3
	Pro Xaa												Xaa	Xaa	Xaa	Xaa	Xaa	25	54
	His Xaa												Xaa	Xaa	Xaa	Xaa	Xaa	25	55
	Pro Xaa												Xaa	Xaa	Xaa	Xaa	Xaa	25	56
Gly	His Xaa	Arg	Val	Val	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	25	57							
Gly	Pro Xaa	Arg	Val	Val	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	25	58							
Gly	His Xaa	Arg	Pro	Leu	Xaa		Xaa	Xaa	Xaa	Xaa	25	59							
Gly	Pro Xaa	Arg	Pro	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	26	60							
Gly	His Xaa	Arg	Val	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	26	61							
Gly	Pro	Arg	Val	Leu	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	20	62							
Gly	His Xaa	Arg	Pro	Val	Xaa	Xaa		Xaa	Xaa		20	63							
Gly		Arg	Pro	Val	Xaa	Xaa		Xaa	Xaa	Xaa	26	64							
Gly	His Xāa	Arg	Val	Val	Xaa	Xaa		Xaa	Xaa		26	65							
_	Pro Xaa	_												Xaa	Xaa	Xaa	Xaa	26	66
	His Xaa														Xaa	Xaa	Xaa	26	67
-	Pro Xaa	_													Xaa	Xaa	Xaa	26	68
	His Xaa														Xaa	Xaa	Xaa	26	69
	Pro Xaa														Xaa	Xaa	Xaa	2	70
-	His Xaa	_													Xaa	Xaa	Xaa	2	71
-	Pro Xaa	-													Xaa	Xaa	Xaa	2	72
	His Xaa														Xaa	Xaa	Xaa	21	73
	Pro Xaa														Xaa	Xaa	Xaa	27	7 4

	_	Per	otid	ев о	E For	mula	a II:	Gly	y-His	s/Pro	-Arc	q-Pro	<b>0/Va</b> ]	<u>-Le</u> ı	ı/Val	L-Xaa	a <sub>2</sub> -Xaa	130
SEQU	ENCE	E								_			_					SEQ ID NO
						Xaa Xaa										Xaa	Xaa	275
						Xaa Xaa										Xaa	Xaa	276
						Xaa Xaa										Xaa	Xaa	277
						Xaa Xaa										Xaa	Xaa	278
						Xaa Xaa										Xaa	Xaa	279
-		_				Xaa Xaa										Xaa	Xaa	280
-		-				Xaa Xaa												281
-		-				Xaa Xaa										Xaa	Xaa	282
-		-				Xaa Xaa											Xaa	283
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_		-				Xaa Xaa											Xaa	285
_		-				Xaa Xaa											Xaa	286
						Xaa Xaa											Xaa	287
_		_				Xaa Xaa											Xaa	288
						Xaa Xaa											Xaa	289
Gly	Pro	Arg	Val	Val	Xaa	Xaa Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	Xaa	290

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37

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<212> TYPE: PRT
<213> ORGANISM: Artificial Sequence
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Glu Arg His Gln Ser Ala Cys Lys Asp Ser Asp Trp Pro Phe Cys Ser
Asp Glu Asp Trp Asn Tyr Lys
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<213> ORGANISM: Artificial Sequence
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<400> SEQUENCE: 293
Gly Pro Arg Val Val Glu Arg His Gln Ser Ala Cys Lys Asp Ser Asp 1 \phantom{-} 15
Trp Pro Phe Cys Ser Asp Glu Asp Trp Asn Tyr Lys 20 25
<210> SEQ ID NO 294
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#### -continued

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<213> ORGANISM: Artificial Sequence
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<400> SEQUENCE: 294
Gly His Arg Pro Leu Asp Lys Lys Arg Glu Glu Ala Pro Ser Leu Arg
Pro Ala Pro Pro Pro Ile Ser Gly Gly Tyr Arg
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<223> OTHER INFORMATION: Xaa=any amino acid
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Xaa Xaa Xaa
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<223> OTHER INFORMATION: Xaa=any amino acid
<400> SEQUENCE: 296
10
Xaa Xaa Xaa
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The invention claimed is:

1. A method of treating inflammation in a subject comprising administering to the subject a peptide

(SEQ ID NO:294)

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Gly His Arg Pro Leu Asp Lys Lys Arg Glu Glu

Ala Pro Ser Leu Arg Pro Ala Pro Pro Pro Ile

Ser Gly Gly Gly Tyr Arg

or a salt or amide thereof, in an amount effective to treat inflammation, wherein the amino terminus is

- wherein R1 and R2 are each selected from the group consisting of hydrogen and a saturated or unsaturated hydrocarbon residue, said residue having from 1 to 10 carbon atoms.
- 2. A method of inhibiting inflammation in a subject comprising administering to the subject a peptide

Gly His Arg Pro Leu Asp Lys Lys Arg Glu Glu

Ala Pro Ser Leu Arg Pro Ala Pro Pro Pro Ile

Ser Gly Gly Gly Tyr Arg

or a salt or amide thereof, in an amount effective to inhibit inflammation, wherein the amino terminus is





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wherein R1 and R2 are each selected from the group consisting of hydrogen and a saturated or unsaturated hydrocarbon residue, said residue having from 1 to 10 carbon atoms.

- 3. The method of claim 1 or 2, wherein the inflammation <sup>5</sup> is due to a condition selected from the group consisting of an infection, an autoimmune condition, a rheumatic disorder, or a disorder of the immune system.
- 4. A method of treating rejection of a transplanted tissue in a subject comprising administering to the subject a <sup>10</sup> peptide

(SEQ ID NO:294) Gly His Arg Pro Leu Asp Lys Lys Arg Glu Glu Ala Pro Ser Leu Arg Pro Ala Pro Pro Pro Ile Ser Gly Gly Gly Tyr Arg 174

or a salt or amide thereof, in an amount effective to treat rejection of transplanted tissue, wherein the amino terminus is



wherein R1 and R2 are either the same or different,

wherein R1 and R2 are each selected from the group consisting of hydrogen and a saturated or unsaturated hydrocarbon residue, said residue having from 1 to 10 carbon atoms.

\* \* \* \* \*